

Effects of insecticide application on *Oracella acuta* (Lobdell) (Homoptera: Pseudococcidae) population and its two dominant parasitoids

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Abstract: The effects of insecticide application on loblolly pine mealybug, *Oracella acuta* (Lobdell) (Homoptera: Pseudococcidae) and its two dominant parasitoids, *Allotropa* sp. (Hymenoptera: Platygasteridae) and *Zarhopalus debarri* Sun (Hymenoptera: Encyrtidae), were investigated using yellow sticky traps. The results revealed that the *Oracella* population responded positively to the intensity of insecticide use, which had adverse effects on its two parasitoids. In fact, the outbreak of the mealybug was largely due to this adverse effect of insecticides on its parasitoids. Orchards with high intensity use of insecticides resulted higher *Oracella* population and *vice versa*. Comparison of three orchards with three levels of insecticide use further demonstrated the effectiveness of the parasitoids in regulating mealybug populations. Significantly more males of *Zarhopalus* sp. were caught than females indicated a sex preference for yellow in this species. A strong positive correlation exists between *Oracella* and its parasitoids but this relationship can be disrupted by the heavy use of insecticides.

Key words: *Oracella acuta*; insecticide; parasite; *Allotropa* sp.; *Zarhopalus debarri*; trapping

The Loblolly pine mealybug, *Oracella acuta* (Lobdell) (Homoptera: Pseudococcidae) is usually only an occasional pest in the southeastern United States. Heavy infestations have occurred only when insecticides have been applied to control cone insects or the Nantucket pine tip moth, *Rhyacionia frustrana*, and are largely due to the disruption of its effective natural enemy complex (Clarke *et al.*, 1990; Sun *et al.*, 1996). There are five primary parasitoids in this complex with two dominant species, *Allotropa* sp. (Hymenoptera: Platygasteridae) and *Zarhopalus debarri* Sun (Hymenoptera: Encyrtidae), which account for about 85% of the total parasitism (unpublished data). Clarke *et al.* (1992) reported the effects of four pyrethroids on four scale insects, including *Oracella acuta*, in a loblolly pine seed orchard in Georgia, and their natural enemies.

Many winged insects are attracted to certain colors

during flight, most commonly to yellow (Jervis and Kidd, 1996). Visual (yellow) sticky traps have been widely used in monitoring some host pest and natural enemy population or in surveys, and in some cases, combined with host's pheromone or semiochemicals to enhance trapping efficiency (Robin and Mitchell, 1987; Samways, 1988; Trimble, 1988; Grout and Richards, 1991; Driesche and Thomas, 1996). However, the efficiency of visual attraction by pests or parasitoids is affected by many factors (Jervis and Kidd, 1996). Yellow traps are particularly efficient in sampling hymenopteran parasitoids such as Ceraphronidae, Selionidae, Platygasteridae, Diapriidae, Mymaridae and Encyrtidae (Masner, 1976; Noyes, 1989).

Since its accidental introduction into Guangdong Province, China in 1988, *Oracella acuta* has spread rapidly and the damage caused by this pest is increasing (Sun *et al.*, 1996). Its distribution increased from

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53.45 km² in 1990 to 355 200 km² in 2001 (Guangdong Forestry Pest Control Station Internal Report). A Sino-US forestry cooperative program was initiated in 1995 to introduce insect natural enemies from the United States to control this pest in China. Results from this on-going project revealed two dominant hymenopteran parasitoids, *Allotropa* sp. and *Z. debarri* that were selected for importation to China (unpublished data).

We conducted this study to further evaluate the effects of insecticide application intensity on *Oracella* populations and those of their parasitoids, and the potential of using yellow, sticky traps as a tool to monitor those parasitoid numbers. The aim of this research is to provide information helpful to the development of a biological control program for *Oracella* in China.

Materials and Methods

Study Site. Three loblolly pine (*Pinus taeda* L.) seed orchards were selected for this study. The South Carolina Orchard (SCN) was located in Jasper County, South Carolina. It lies about 130 km east of Lyons Orchard (LO), which is located in Toombs County, Georgia. The third orchard, the Bowater Orchard (BOW), is also located in Toombs County, Georgia, about 16 km north of Lyons Orchard. We only used a portion of each orchard as our study site. Trees in those portions were all around 10 years old and the same size. We classified these three sites as heavily sprayed (BOW), lightly sprayed (SCN) and moderately sprayed (LO). Thus, the three sites corresponded to three treatments with respect to insecticide application intensity (Table 1).

Data Collection. On each site, three Pherocon[®] Adult Monitoring (AM-N/B) Traps, a folded, double-layer, yellow, sticky trap (14 cm × 23 cm) with a pre-punch locking tab and hanger hole, were suspended on low branches on the edge of the crowns of individual trees. The traps were about 2 m above the ground and were about 50 m apart in a triangular pattern. The traps were set out on April 1 and remained until October 30 each year on each site. The traps were changed a total of 10 times at 2 – 4 weeks intervals. On each trap replacement date, collected traps were taken back to the labo-

ratory and checked under a dissecting microscope, male *Oracella* (both apterous and alate males) and two parasitoids (*Allotropa* sp. and *Z. debarri*) were counted and recorded. In some cases, Pure Lemon (Sysco) solvent was required to remove parasitoids from the trap for observation and subsequent identification and sexing. Female parasitoids were also dissected and observed under a microscope to determine their maturity (ovary observation) but male sexual maturity was not determined in this study.

Table 1 Insecticides and aerial spray date (day/month) at three orchards in 1996 and 1997

	SCN	LO	BOW
1996	Bifenthrin, 21/7	Azinphosmethyl, 19/4	Azinphosmethyl, 13/4
		Azinphosmethyl, 1/6	Azinphosmethyl, 29/4
	Bifenthrin, 23/8	Azinphosmethyl, 2/7	Azinphosmethyl, 1/6
		Asana xl, 23/8	Azinphosmethyl, 17/7
1997	Bifenthrin, 17/3	Asana xl, 23/8	Azinphosmethyl, 30/8
			Azinphosmethyl, 20/3
			Azinphosmethyl, 3/4
			Azinphosmethyl, 2/5
			Azinphosmethyl, 30/6
			Azinphosmethyl, 17/8
			Azinphosmethyl, 29/10

At the beginning of each field season, 100 shoots were randomly cut from each site. Wax cells and parasitized mummies were counted for comparison with early trapping data.

Total catches of both *Oracella* males and the two parasitoid species, and shoot dissection data were analyzed by analysis of variance and means were separated by the Sidak *t* test (SAS Institute, 1989). The paired-*t* test was used to test for sexual differences in catches of the two parasitoid species. Regression analysis was used to determine the relationships between *Oracella* and the parasitoids due to insecticide application.

Results and Discussion

Insecticide impact on the malybug. There were significant differences in total mealybug catches among the three orchards ($F = 101.97$, $df = 2$, $P = 0.05$).

BOW had the highest mealybug population, followed by LO and SCN. This result corresponds to the intensity of insecticide application at each orchard (Table 1). The general theory regarding *Oracella* outbreaks is associated with insecticide application (Clarke *et al.*, 1990, 1992, Sun *et al.*, 1996). This result clearly supports this hypothesis. To some degree, the *Oracella* population seems to positively respond to insecticide application. The same trend was observed over the whole trapping season (Table 2). This result was very similar to the data obtained by systematically sampling shoots and counting wax cells in 1995 and 1996 (unpublished data) and result by Clarke *et al.* (1990). Early generation (overwintering) accounts for the majority of the whole season population of *Oracella* (Xu *et al.*, 1992). In BOW, the *Oracella* population remained consistently high until October when mealybugs prepared to overwinter, largely because insecticides were applied 5 times in 1996 and 6 times in 1997. In this case we expected a high *Oracella* population early the following year. However, if insecticide use was stopped, its population size would be expected to be more or less similar to that in LO or SCN since parasitoids are known to bring *Oracella* populations down (unpublished data). In LO and especially SCN, insecticides were applied twice in 1996 and once and twice respectively in 1997. The effects of insecticides on parasitoids at SCN in 1997 were negligible since it was sprayed in mid-March when most parasitoids are not active. As we can see from Table 2, the *Oracella* population declined rapidly after the overwintering generation, and then basically remained constant. Field observations confirmed this, high level of wax cells per shoot were observed in BOW over the whole season but only in April in LO and SCN (the lowest population level), and wax cells, although present, were very hard to find. These scenarios clearly demonstrate the effectiveness of the parasitoids. There was a significant difference between LO and SCN in April – June data, but there was no significant difference in the rest of the year, even though traps in LO still caught more mealybugs than in SCN. Clearly, high intensity application of insecticides appears to result in high mealybug popula-

tions, moderate intensity of insecticide application in moderate mealybug populations, and low insecticide use in low mealybug populations. These results indirectly reflect the effectiveness of the parasitoids in controlling mealybugs.

Further evidence to support this effect was provided by shoot dissection in April, 1997 (Table 3). Significant differences were found among the three orchards (Sidak *t* test, $df = 297$, $t = 2.40$, $P = 0.05$). BOW had the highest number of wax cells per shoot, followed by SCN and LO, which were not statistically different. In the case of LO, we expected the same cycle in 1998 as in 1997 since one application was conducted in August. But, there were no noticeable wax cells observed in 1998 which was confirmed by attempted sampling in 1998 for parasitoid shipment to China. SCN would be expected to have returned to the natural stand situation where no wax cells can be found.

Insecticide impact on parasitoids. For *Z. debarri*, overall catches per trap in SCN (41.8) was significantly higher than both in LO (20.4) and BOW (20.3) ($F = 10.7$, $df = 2$, $P = 0.05$). But there was no significant difference between LO and SCN in overall catches. Not surprisingly, nearly all catches were recorded in early spring in SCN (Table 2). One reason for this is that the *Z. debarri* population reaches its peak in early spring (unpublished data). Secondly SCN had more or less approached a natural stand situation after two applications of insecticide in 1996 which led to a reasonable *Oracella* population in early 1997 followed by a rise in parasitoid numbers which quickly brought the *Oracella* population down to a natural stand level. This was followed by decline of the parasitoid. Once again, this demonstrates the effectiveness of the parasitoid as a biocontrol agent for mealybugs. In both LO and SCN, there was no clear pattern observed, and *Z. debarri* population fluctuated. In our previous study (unpublished data), *Z. debarri* usually has two population peaks, one in early spring and one in fall. This still seems the case with one peak in April and one in August (Table 2), but this trend could also be altered by insecticide applications in the orchard, directly by killing the parasitoid, or indirectly by affecting its host

Table 2 Mean ($\bar{X} \pm SD$) catches/per trap of male *Oracella acuta*, *Allotropa* sp. and *Zarhopalus debarri* in three orchards

SITE	COLLECTION DATE										
	1 APRIL	15 APRIL	29 APRIL	2 JUNE	18 JUNE	9 JULY	13 AUG.	9 SEPT.	29 SEPT.	15 OCT.	30 OCT.
Mean catches of <i>Oracella</i> male per trap											
BOW	247.3 ± 54.0 a	394.3 ± 826.6 a	1053.7 ± 48.3 a	414.7 ± 105.6 a	243.0 ± 255.6 a	146.7 ± 61.9 a	351.3 ± 144.3 a	376.7 ± 187.3 a	80.0 ± 42.9 a	8.0 ± 9.5 a	3 ± 1.5 a
LO	128.0 ± 29.5 b	190.0 ± 47.8 b	625.0 ± 164.5 b	105.3 ± 26.6 b	61.3 ± 11.5 b	54.7 ± 30.7 b	94.3 ± 16.3 b	91.7 ± 30.4 b	40.7 ± 9.3 b	7.3 ± 1.5 a	3 ± 2.3 a
SCN	104.7 ± 32.9 b	69.3 ± 29.9 c	149.0 ± 7.0 c	28.7 ± 16.7 c	40.0 ± 21.1 b	18.0 ± 7.9 b	20.7 ± 6.1 b	16.0 ± 8.7 b	13.0 ± 2.0 b	5.3 ± 1.5 a	2 ± 2.7 a
Mean catches of <i>Allotropa</i> sp. per trap											
BOW	3.0 ± 2.6 a	4.7 ± 2.1 a	1.7 ± 0.6 a	6.3 ± 2.1 a	6.0 ± 1.0 a	5.0 ± 1.7 a	6.0 ± 2.6 a	1.7 ± 1.0 a	1.0 ± 1.0 a	1.0 ± 1.7 a	0.0 ± 0.0 a
LO	3.0 ± 1.7 a	4.0 ± 1.0 a	1.7 ± 0.6 a	2.0 ± 0.0 b	3.7 ± 1.2 a	2.3 ± 0.6 b	3.3 ± 1.5 b	0.3 ± 0.6 a	0.3 ± 0.6 a	0.7 ± 0.6 a	0.0 ± 0.0 a
SCN	10.7 ± 3.5 b	5.0 ± 3.0 a	4.7 ± 3.2 b	1.7 ± 0.6 b	4.0 ± 1.0 a	2.7 ± 1.5 bc	2.0 ± 0.0 b	1.3 ± 1.2 a	1.0 ± 1.0 a	0.3 ± 0.6 a	0.0 ± 0.0 a
Mean catches of <i>Zarhop alius deharri</i> per trap											
BOW	0.7 ± 0.6 a	1.7 ± 0.6 a	4.3 ± 1.2 a	1.0 ± 1.0 a	1.7 ± 1.5 a	2.7 ± 1.2 a	6.3 ± 1.5 a	1.3 ± 0.6 a	1.3 ± 0.6 a	0.0 ± 0.0 a	0.0 ± 0.0 a
LO	2.7 ± 2.5 b	2.7 ± 1.2 a	3.0 ± 1.7 a	0.3 ± 0.6 a	0.7 ± 0.6 a	1.3 ± 1.5 a	6.3 ± 1.2 a	1.7 ± 0.6 a	1.7 ± 0.6 a	0.0 ± 0.0 a	0.0 ± 0.0 a
SCN	3.3 ± 0.6 b	13.0 ± 10.4 b	21.3 ± 2.1 b	1.3 ± 1.5 a	1.7 ± 0.6 a	0.3 ± 0.6 a	0.3 ± 0.6 a	0.3 ± 0.6 a	0.3 ± 0.6 a	0.0 ± 0.0 a	0.0 ± 0.0 a
Mean ratio of the two parasitoids/ <i>Oracella</i>											
BOW	1.5 ± 0.7 a	1.6 ± 0.4 a	0.6 ± 0.2 a	1.9 ± 0.7 a	5.7 ± 3.9 a	5.8 ± 2.3 a	2.7 ± 0.9 a	2.6 ± 1.5 a	2.7 ± 0.4 a	5.3 ± 9.1 a	5.0 ± 3.0 a
LO	4.2 ± 2.7 a	3.5 ± 0.6 a	0.8 ± 0.4 a	2.3 ± 1.2 a	7.0 ± 1.8 a	6.7 ± 4.6 a	4.7 ± 1.8 a	7.5 ± 0.9 a	4.8 ± 1.5 a	8.5 ± 7.5 a	4.0 ± 1.3 a
SCN	14.6 ± 6.5 b	27.8 ± 10.3 b	17.5 ± 4.2 b	16.8 ± 9.0 b	18.7 ± 14.3 b	20.7 ± 13.4 b	11.6 ± 1.8 a	9.9 ± 9.2 a	10.4 ± 4.5 a	4.8 ± 8.2 a	3.5 ± 2.0 a

Means in a column followed by the same letter are not significantly different (Sidak *t* test, $P = 0.05$).

mealybug population. It seems that the parasitoids respond to insecticide application more rapidly than mealybugs (Table 1 and 2), probably due to the mealybugs wax cells providing better protection against insecticides. This explanation should be viewed with caution because not only the application times, but also the insecticides and application rate all greatly affect parasitoid numbers.

More *Allotropa* sp. were caught in SCN than at both BOW and LO, especially in early April when there was a reasonably stable population of mealybugs in SCN. With the decline of the mealybug population the *Allotropa* sp. population also declined (Table 2). There is a significant difference in overall mean catches per trap among the three orchards ($F = 7.21$, $df = 2$, $P = 0.05$). *Allotropa* sp. is a parasitoid present all year around that is most abundant during summer. This was clearly the case in BOW and LO but not in SCN for the reason mentioned above. The effects of each pesticide application on this parasitoid are unclear, perhaps due to the fact that *Allotropa* sp. parasitizes all stages of the mealybug except eggs. With the overlapping generations that occur over summer there would always be a certain number of mummies present which are less susceptible to insecticides than emerged adult parasitoids. Some mummies are protected in mealybug wax cells. So only those adult parasitoids or parasitized mealybug crawlers are likely to be susceptible to sprays. In this respect, *Z. debarri* are less susceptible to insecticides than *Allotropa* sp. considering that most *Z. debarri* appear in early spring or fall and parasitize pre-adult female mealybugs that are protected by wax cells.

We caught significantly more *Allotropa* sp. than *Z. debarri* in BOW during the whole season (Paired t -test, $n = 30$, $t = 2$, $P = 0.047$). This conforms the composition of the mealybug parasitoid complex with *Allotropa* sp. being the most abundant, followed by *Z. debarri* (unpublished data). The degree of this dominance among the three orchards can vary due to insecticide effects on parasitoids. For example, intensive spraying during the summer could reduce *Allotropa* sp. populations greatly in BOW, resulting in lower capture rates than it would otherwise have occurred. The attrac-

tiveness of yellow traps to individual parasitoids depends, to some extent, on the latter's physiological condition (Jervis and Kidd, 1996). Schneider (1969) suggested that color preference by parasitoids may be influenced by the color of the most abundant flowers in bloom at the time of sampling. Obviously, great caution must be used when estimating the relative abundance of the two parasitoid species from yellow sticky traps. Dissection of some parasitoids in the laboratory revealed that most females caught in traps were not sexually mature, which suggests immature parasitoids are most attracted to yellow trapping. A similar result was reported in a study on cone flies (Roques *et al.*, 1995). A few more *Allotropa* sp. were caught in LO, but this was not significant (Paired t -test, $n = 30$, $t = 0.40$, $P = 0.69$), probably because its low level *Oracella* population was unable to sustain a reasonable *Allotropa* sp. population. Not surprisingly, we caught more *Z. debarri* than *Allotropa* sp. in SCN even though this was not significant (Paired t test, $n = 30$, $t = 77$, $P = 0.45$). This was because most captures were in early spring which was dominated by *Z. debarri*, after which both *Oracella* and the parasitoid populations collapsed.

In terms of sex-specific response to yellow sticky traps, for *Allotropa* sp., we trapped significantly more females than males in LO (Paired t -test, $n = 30$, $t = 2.82$, $P = 0.086$). More females were trapped in BOW and SCN but this was not statistically significant, with 0.07 times more females in SCN (Paired t -test, $n = 30$, $t = 1.83$, $P = 0.085$) and 0.63 times more female in BOW (Paired t -test, $n = 30$, $t = 1.83$, $P = 0.08$). The natural sex ratio ($\text{♀}/\text{♂}$) for *Allotropa* sp. is 1.11 (unpublished data). So with certainty, we can say that the sex ratio for *Allotropa* sp. in SCN, which was closest to a natural situation, remained normal. In both Bow and LO, more females were trapped than would be predicted by the natural sex ratio. This difference could be due to insecticide disruption or sexual bias in response to the yellow trap color, which is usually related to host detection or flower colors on which the parasitoid feeds (Driesches and Bellows, 1996).

For *Z. debarri*, significantly more males were caught than females in all three orchards, Bow (Paired

t-test, $n = 30$, $t = 4.71$, $P = 0.0001$), LO (Paired *t*-test, $n = 30$, $t = 2.62$, $P = 0.014$) and SCN (Paired *t*-test, $n = 30$, $t = 2.62$, $P = 0.014$). The natural sex ratio ($\text{♀}/\text{♂}$) for *Z. debarri* is 1.43 (Sun *et al.*, 1998). This strong sex-specific response to yellow traps is most likely due to the preference of male *Z. debarri* for yellow. However, the mechanism underlying this preference remains unknown. Both male and female *Z. debarri* have supplementary food sources. This difference could reflect difference in foraging pattern, but more likely it may suggest that yellow may represent a behavioral signal for males. As reported, some parasitoids use visual cues such as color in host habitat location (Jervis and Kidd, 1996) or a similarity between traps and host plants provides some stimulus for parasitoids (Driesches and Bellows, 1996).

Parasitoids/*Oracella* ratio. The parasitoids, *Allotropa* sp. and *Z. debarri* together account for about 85% of all *Oracella* parasitism (unpublished data). Therefore, we can presume that the trap catch ratio of the two parasitoids/*Oracella* represents the trend in the parasitism rate. Overall there was a significant difference between BOW, SCN and LO ($F = 38.29$, $df = 2$, $P = 0.050$), but there was no significant difference between LO and SCN. The ratio in SCN remained relative high until August, even after both *Oracella* and the parasitoid populations had collapsed after spring. Exactly as expected, the ratio in BOW was very low, but the low ratio in LO compared to SCN was unexpected (Table 2). In heavily sprayed BOW, and to a lesser extent LO, parasitism was highest during June and July. A similar result was reported by Clarke *et al.* (1990), which indirectly supports the dominant status of *Allotropa* sp.

A better result was obtained from shoot dissection with real parasitism (Table 3). There was no significant difference between SCN and LO but both were significantly higher than BOW. The low ratio in trap catches relative to shoot dissection indicated that trap catches reflected a general trend, but not real parasitism. As discussed earlier, many factors affect catches, such as attractiveness of yellow traps, hyperparasitism, *etc.* We noticed a significant number of *Chartocerus* sp. in

traps, a hyperparasite present in LO. Why more *Chartocerus* sp. were presented in LO remains unknown to us. Also, we did not detect the other three minor parasitoids (*Acerophagus coccis*, *Aprostocetus* sp. and *Aenasius* sp.) in trap catches.

Table 3 Mean ($\bar{X} \pm SD$) of *Oracella* density (wax cells) per shoot and parasitism at three seed orchards

Site	Shoots dissected	<i>Oracella</i> density	Parasitism (%)
BOW	100	6.7 ± 3.4 a	3.3 ± 1.1 a
LO	100	1.92 ± 1.0 b	26.9 ± 7.4 b
SCN	100	2.7 ± 1.5 b	35.9 ± 9.7 b

Means in a column followed by the same letter are not significant difference ($P = 0.05$, Sidak *t* test.)

Linear regression analysis showed that there was a strong correlation between *Oracella* catches and *Allotropa* sp. catches ($R^2 = 0.73$, $P = 0.01$) in SCN, but not in BOW ($P = 0.73$) and LO ($P = 0.15$). A less strong relationship was found for *Z. debarri* ($R^2 = 0.42$, $P = 0.21$) in SCN, BOW ($P = 0.67$) and LO ($P = 0.60$). This is largely due to that fact that *Z. debarri* is a seasonal species in spring and fall while *Allotropa* sp. is present year around with a peak of abundance in summer. As a result, a strong relation exists between the ratio of the two parasitoids/*Oracella* and *Oracella* catches in SCN only ($R^2 = 0.84$, $P = 0.001$). This result demonstrates that a close relationship exists between those two parasitoids and their host in a natural situation, and that insecticide application can disrupt this relationship and the balance between them causing outbreaks of the mealybug.

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杀虫剂对湿地松粉蚧种群及其天敌的影响

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摘要: 湿地松粉蚧是于 1988 年传入我国广东省的一种重要林业外来入侵害虫。现在该害虫在我国的分布面积为 35.52 万公顷, 严重影响着我国南方松林的生长健康。该害虫在其原产地美国南方并不造成大的危害, 也不是一种主要害虫。只有当大量应用杀虫剂防治其它害虫时, 由于杀死了其天敌, 湿地松粉蚧种群才会明显增长。为控制这一外来入侵害虫, 中美两国于 1995 年开展了从美国引进天敌防治广东省湿地松粉蚧的林业合作项目。本文报道了 1996~1997 年间在美国南方三个种子园使用杀虫剂防治球果种实害虫时, 杀虫剂对湿地松粉蚧种群及其两种主要天敌有明显的影 响, 这也间接地说明了寄生性天敌对湿地松粉蚧在自然条件下的控制作用。相关分析显示湿地松粉蚧种群数量与其天敌是密切相关的, 但杀虫剂可以打破这种平衡。这一方面说明从美国引进天敌防治湿地松粉蚧是可行的, 另一方面也显示在美国采集湿地松粉蚧天敌应在使用过杀虫剂后的林分中。

关键词: 湿地松粉蚧; 杀虫剂; 天敌诱捕; 广腹细蜂; 迪氏跳小蜂; 引诱

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